essential component for the viral replication as well as an attractive target for antiviral drug development. The specificity of NV Pro depends on the primary sequence of the cleavage site where the Q/G at P1/P1' position of a polyprotein peptide is preferentially cleaved followed by E/A or E/G as a secondary sites. X-ray crystallographic studies also show that NV Pro adopts a serine protease-like fold containing two β-barrel domains separated by a cleft within which lie the active-site catalytic residues. In addition to the active site, it was suggested that mutual conformational rearrangement of Pro/peptide binding site plays a pivotal and common role in efficient substrate recognition of Pro from different genotypes, GI and GII. These findings lead us toward the development of substrate-based peptidemimetic inhibitors with broad-spectrum activity. We are using solution NMR spectroscopy to solve the solution structure of NV Pro and investigate the interaction mode of NV Pro and inhibitor(s) in parallel with in vitro assay using a fluorescent substrate derived from the natural cleavage site of NV. Here we will present the solution structure of NV Pro solved by NMR spectroscopy. Preliminary results on structural and dynamical characterization of the protease-inhibitor interaction will also be presented.

3275-Pos Board B380
Structural Characterization of the Zinc Finger Domain of Cytoplasmic Polyadenylation Element-Binding Protein
Daniel Merkel, Bryce Hilburn, Sarah Wells, Stephanie Geiser, Haley Hoover, Oluwatobi Ajoku, Brian Lee.
Cytoplasmic polyadenylation element-binding protein (CPEB) is an important factor in translational regulation of oogenesis, cellular senescence and synaptic plasticity. It does this by regulating a poly(A) tail elongation through interactions with a number of other proteins. The region of mRNA that CPEB binds to is a uracil rich region known as the cytoplasmic polyadenylation element (CPE). The carboxy terminal region of CPEB is composed of three regions that are required for CPE recognition and binding within the 3' UTR of mRNA. It has two RNA binding domains and a zinc finger motif. The zinc finger region contains six cysteine and two histidine amino acids that are highly conserved throughout many species. The conserved residues suggest the presence of a zinc finger structure containing two zinc ions. The zinc finger region of CPEB does not seem to be homologous with any know zinc fingers. We are currently in the process of structural characterization using NMR techniques.

3276-Pos Board B381
Domain Structure of the Major Allergen Ovomucoid by Solution NMR
Natalie E. Stenzoski.
Domain Structure of the Major Allergen Ovomucoid by Solution NMR
Natalie E. Stenzoski, Gugsa W. Gebriel, Bruce D. Ray, Horia I. Petrache
The interest in the ovomucoid protein is twofold. First it is a protein of interest for medical studies due to its potent allergen activity. Second, as a special variety of glycosylated protein (Kazal family), it allows us to explore the role of protein glycosylation for a particular or model case. Glycosylated proteins are commonly secreted by tissues as signaling agents. For example, the serpin family serine proteinase inhibitor, α-1 proteinase inhibitor (A1AT/ α-1 antitrypsin) regulates trypsin. Recent research in our laboratory indicates that A1AT interacts with lipid membranes and affects ionic currents through the membrane. In contrast, ovomucoids do not. This difference in membrane interaction is expected to have a structural cause. However, very little overall structural data is available. The nature, location, and orientation of the glycosyl groups are determining factors in protein-membrane interactions and are deeply involved biological effects of glycosylated proteins, including effects on ion transport. We use solution NMR spectroscopy to determine the structure of the chicken ovomucoid protein, taking advantage of the division of its structure into three stable domains of 55-65 amino acids each. We present results on the protein purification steps and isolation of separate domains and the models of individual domains and overall protein structure from the analysis of NMR spectra.

3277-Pos Board B382
Solution Structure of a Central Domain of the Conjugative Transfer Protein TraI
Nathan T. Wright, Joel F. Schildbach.
TraI, the F plasmid-encoded nickase, is a 1756 amino acid protein essential for conjugative transfer of F plasmid DNA from one bacterium to another. While crystal structures of N- and C-terminal domains of F TraI have been determined, central domains of the protein are structurally unexplored. These middle domains (between residues 306 and 1500) are known to both bind single-stranded DNA (ssDNA) and unwind DNA through a highly processive helicase activity. Of this central region, the more C-terminal portion (~900-1500) appears related to helicase RecD of the E. coli RecBCD complex. The more N-terminal portion (306-900), however, shows limited sequence similarity to other proteins. In an attempt to define the structure of well-folded domains of this middle region and discern their function, we have isolated stable regions of TraI following limited proteolysis. One of these regions, TraI (381-569), was identified and shown to be well-folded in solution via NMR. Here, we present the high resolution solution structure of this region. We show that, like the 900-1500 region, 381-569 is part of a RecD-like fold. We also show where the 381-569 region is located relative to the rest of the protein via a series of SANS and SAXS experiments. Together, these data provide structural explanations to several questions about the exact mechanism of TraI-dependant plasmid transfer.

3278-Pos Board B383
Application of Magnetic Resonance for Metabolomic Investigation of Mollusks
Andrey P. Tikunov, Haakil Lee, Michael K. Stoskopf, Jeffrey M. Macdonald.
Metabolites, as the end products of metabolism, represent the functional responses of a cell. Their characterization can provide insight into the underlying mechanisms of genomic or environmental actions on metabolism. The aquatic environment is varied and dynamic, providing a vast diversity of physical and chemical challenges to metabolism, making the study of the metabolites of mollusks particularly fruitful for scientists interested in comparative physiology, pharmacology and toxicology. Organ specific metabolic fingerprints can establish time dependent assessments for interpreting functional adaptations to environmental and nutritional challenges using either invasive tissue extraction from multiple individuals or non-invasive longitudinal observation of the same individual. NMR spectroscopy and magnetic resonance imaging (MRI) permit non-invasive monitoring of the metabolome.
The mollusks are a useful, robust model organism for tissue metabolism studies. Its relatively few organs are easily delineated and there is sufficient understanding of their functions based on classical assays to support interpretation of advanced spectroscopic approaches. Here we apply high-resolution proton and carbon nuclear magnetic resonance (1H and 13C NMR)-based metabolomic analysis to Eastern oyster C. virginica and freshwater mussel Eastern Elliptio E. Complanata to investigate the variation in the metabolic profile of different organ groups. MRI was used to non-invasively identify the morphology of the organs. In vivo MR spectra can be obtained from single region of interest (ROI or voxel) or multiple ROI simultaneously using the technique typically called chemical shift imaging (CSI). Here we also report applications of CSI to marine samples and describe the use of the technique to study in vivo glycine metabolism in oysters using 13C MRS.
A biochemical schematic is presented that relates metabolites to biochemical pathways correlated with physiological organ functions.

3279-Pos Board B384
Paramagnetic Contributions to Nuclear Spin-Lattice Relaxation in Proteins
Robert G. Bryant, Galina Diakova, Yanina Goddard, Jean-Pierre Korb.
Paramagnetic contributions to spin-lattice relaxation rate constants are very useful in providing long-range distance constraints for structural determinations by NMR and also for controlling image contrast in clinical MRI protocols. When a radical with a long electron-spin-relaxation time is bound to a protein, the paramagnetic contribution to the water-proton-spin-lattice-relaxation rate is larger than predicted by usual theory. We show that this excess nuclear spin relaxation efficiency results from long-lived-bound water molecules for which the electron-nuclear coupling is correlated with the long rotational correlation time of the protein. Because the correlation time is so long compared with the relative translational correlation times, even distant water molecules make significant contributions and increase the relaxation efficiency of the paramagnetic center. If the rotational motion of the protein is stopped, as in a solid protein, a protein gel, or a tissue, the paramagnetic contributions to relaxation are both qualitatively and quantitatively changed from the solution limit. The spin-lattice relaxation for different paramagnetic systems may be understood in terms of the intrinsic dynamics of the protein and the spin dynamics of the paramagnetic center, which depends on the identity and magnetic characteristics of the paramagnetic center. These different effects are important for applications to magnetic imaging and structure determination by solid state NMR.